The Interface Between Kinetics and Dynamics: Physiological Modeling of Two Estrogen Receptor Agonists

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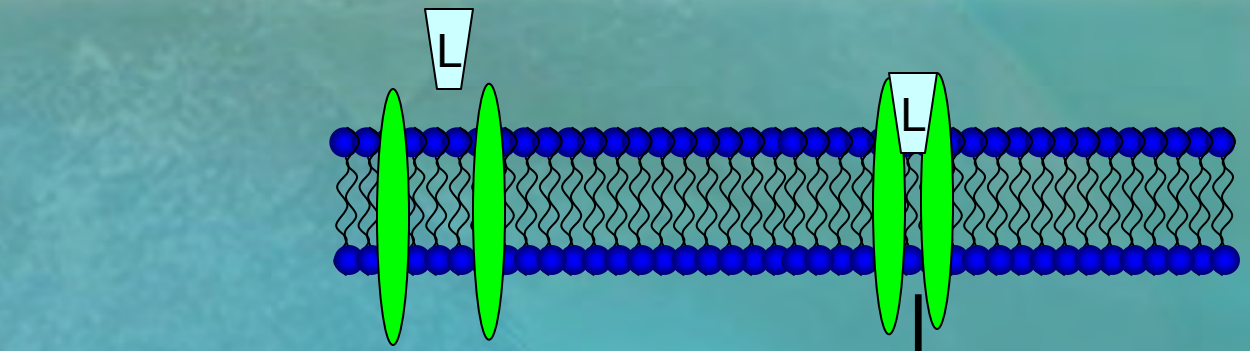
West to East with Paul West

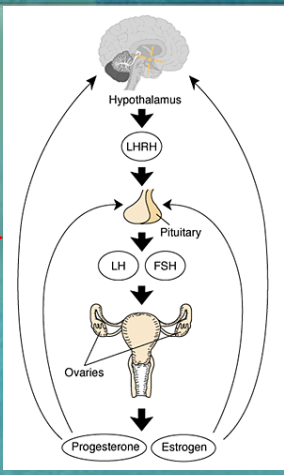
Much of today's public anxiety about science is that apprehension that we may be forever overlooking the whole by an endless, obsessive preoccupation with the parts.

Lewis Thomas

Computational Toxicology!

The Pharmacokinetic Pharmacodynamic Interface





K_{abs}

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Organism ADME

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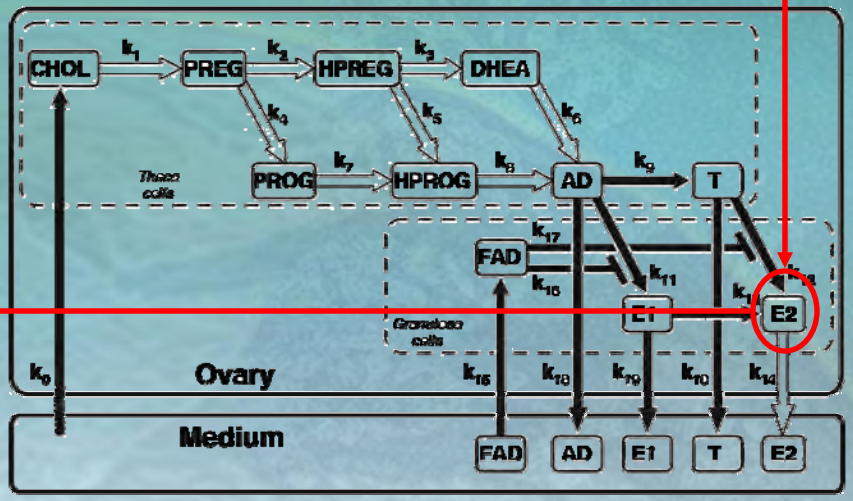
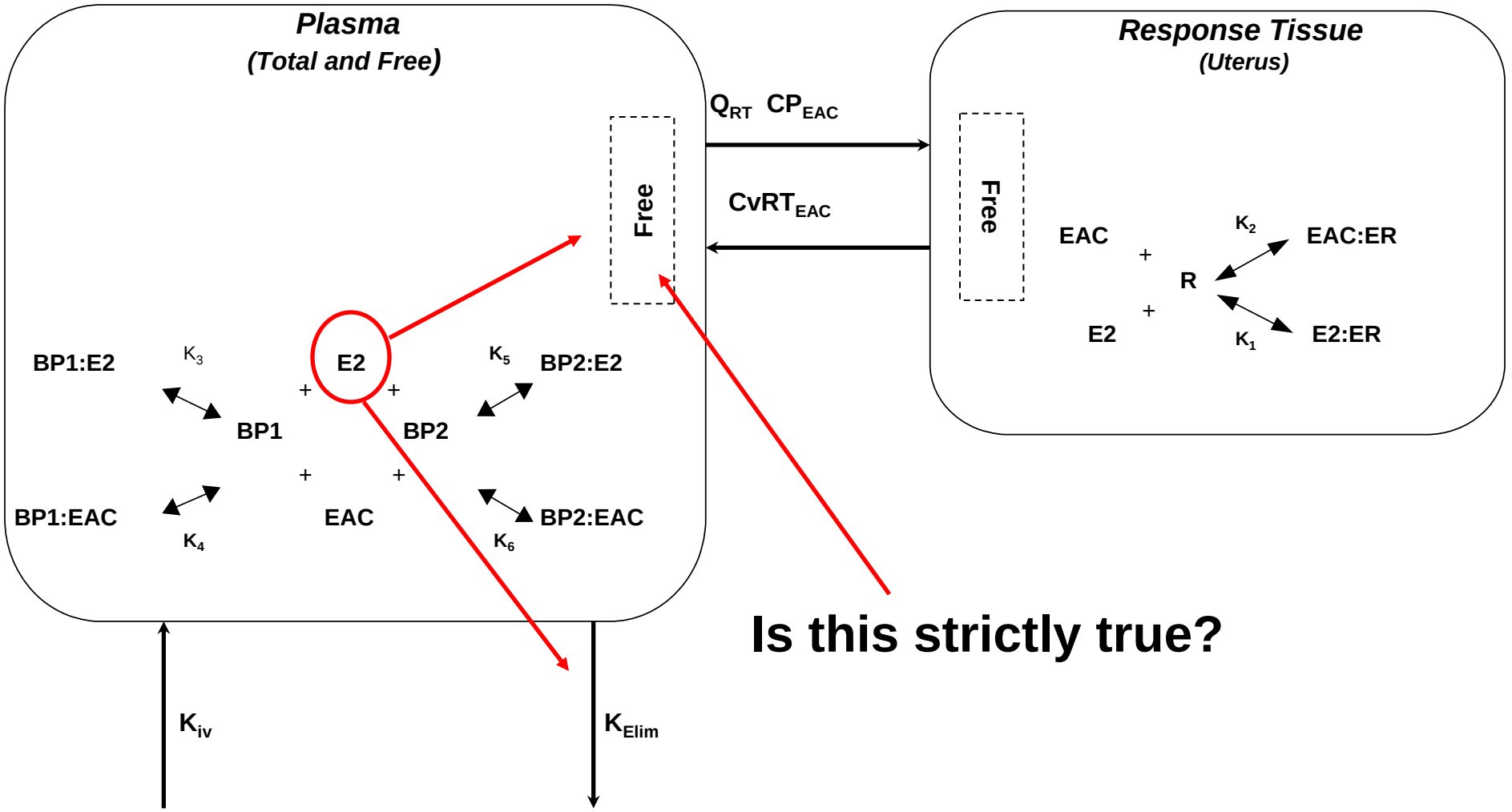
Tissue Receptor Occupancy

Tissue Dynamics



Breen et al. 2007

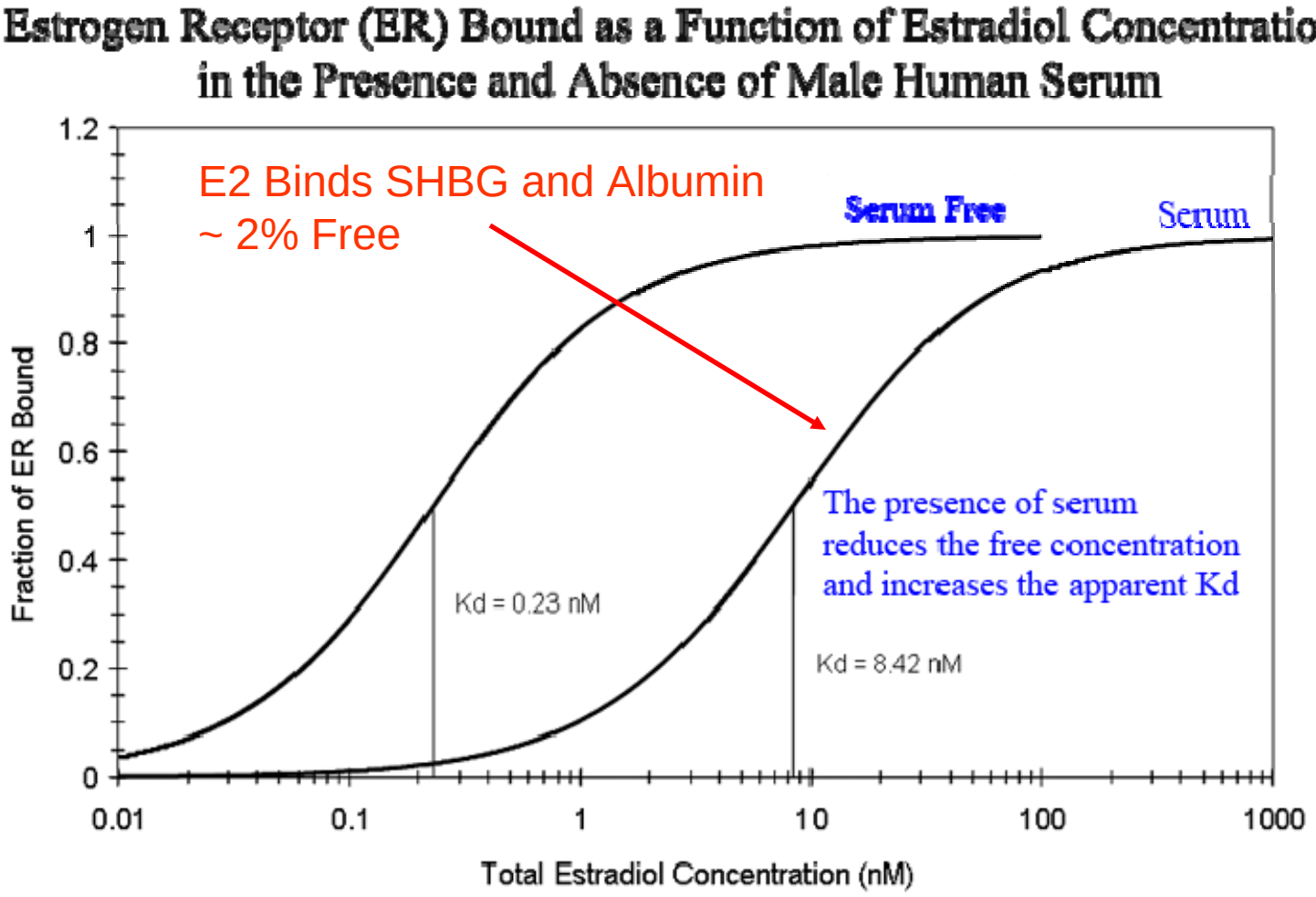
Integrating Protein Binding, Receptor Binding and Clearance Using *In Vivo* Models



Do Saturable Binding Processes Affect Dosimetry?

- **Is response ~ receptor occupancy?**
- **Do the multiple binding equilibria affect dosimetry? Under what conditions?**
- **Does protein binding affect clearance and potency?**

Serum Protein Binding Restricts Availability



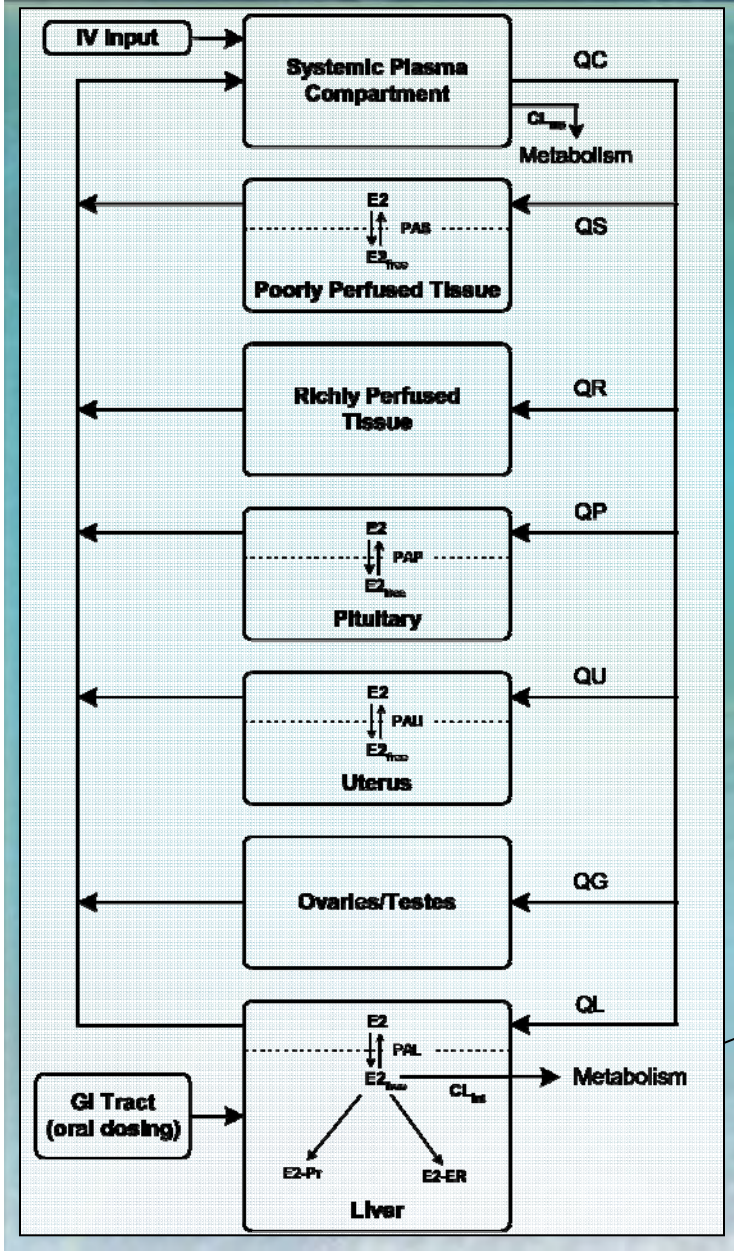
Adapted from: Teeguarden, J. G., and Barton, H. A. (2004). *Risk Anal* **24**, 751-70.

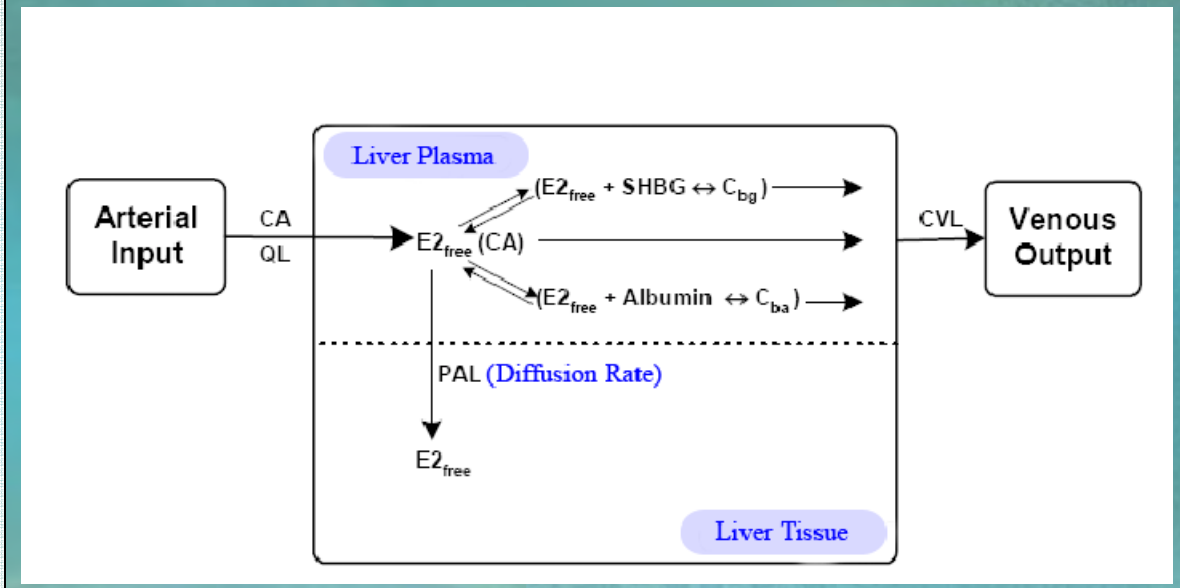
Clearance Affects Potency

Compound	Potency Relative to E2		
	- Clearance		+ Clearance
	No Serum	Rat Serum	Rat Serum
Estradiol	1	1	1
Genistein	0.02	0.16	0.32
Bisphenol A	0.0005	0.0009	0.002

Estradiol Pharmacokinetics and Receptor Binding Model

Estradiol PBPK Model

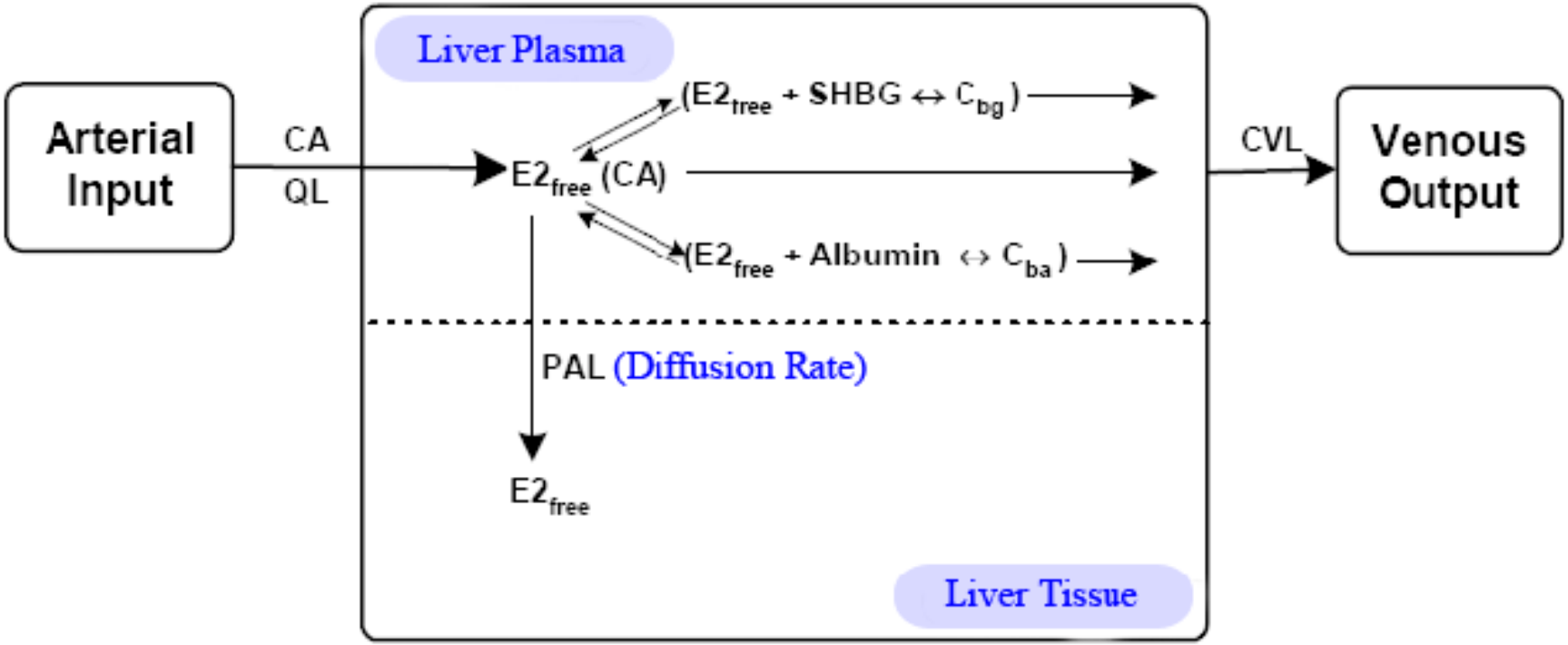




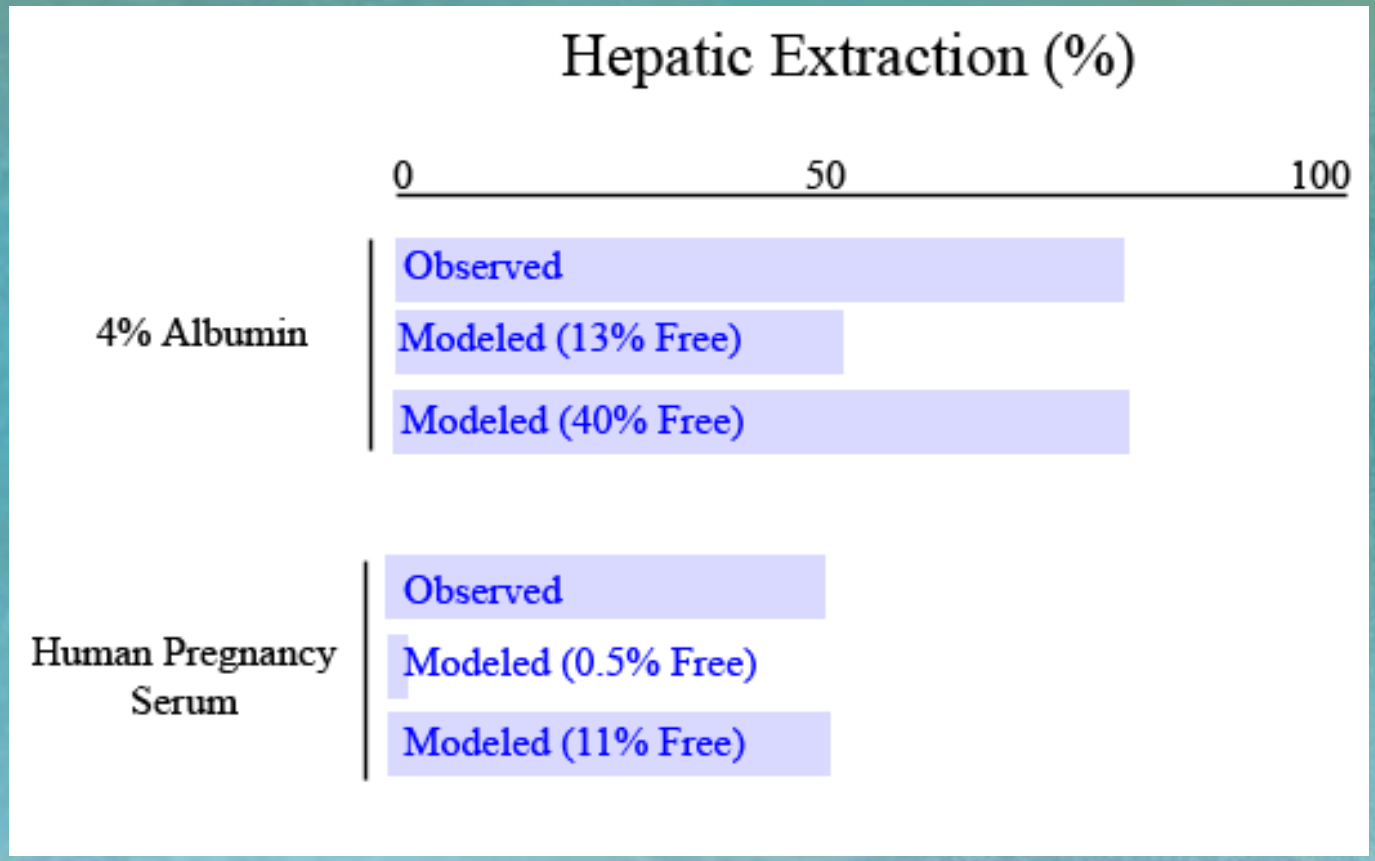
Hepatic Extraction Model

Does Binding Affect Hepatic Clearance?

Isolated perfused liver model

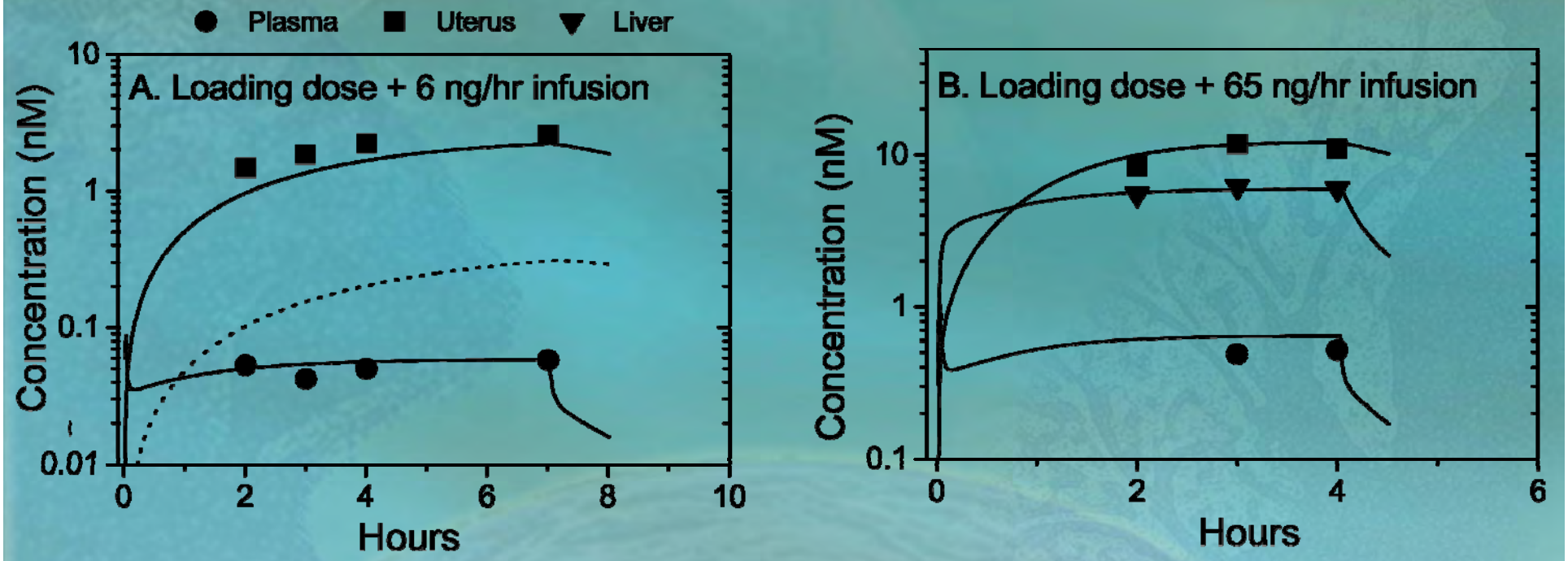


Yes, Binding Affects Clearance



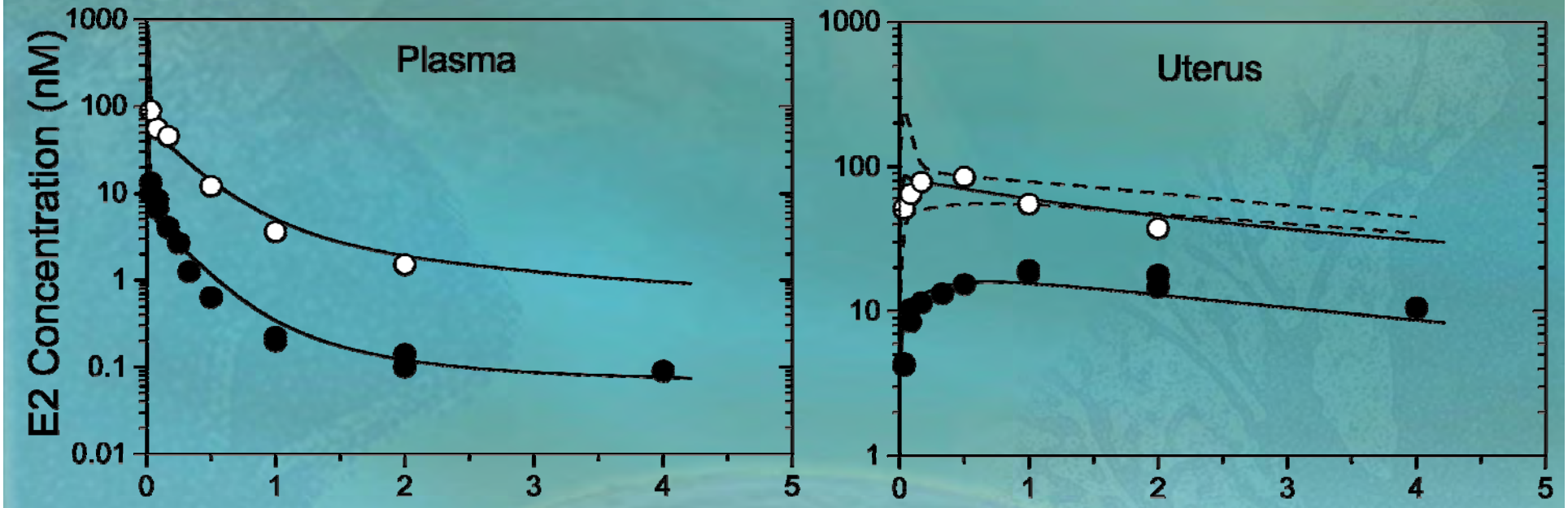
- **Disassociation of estradiol during transit through the liver increases the "available" fraction of estradiol for diffusion and metabolism**

E2 Pharmacokinetics in Intact Female Rats



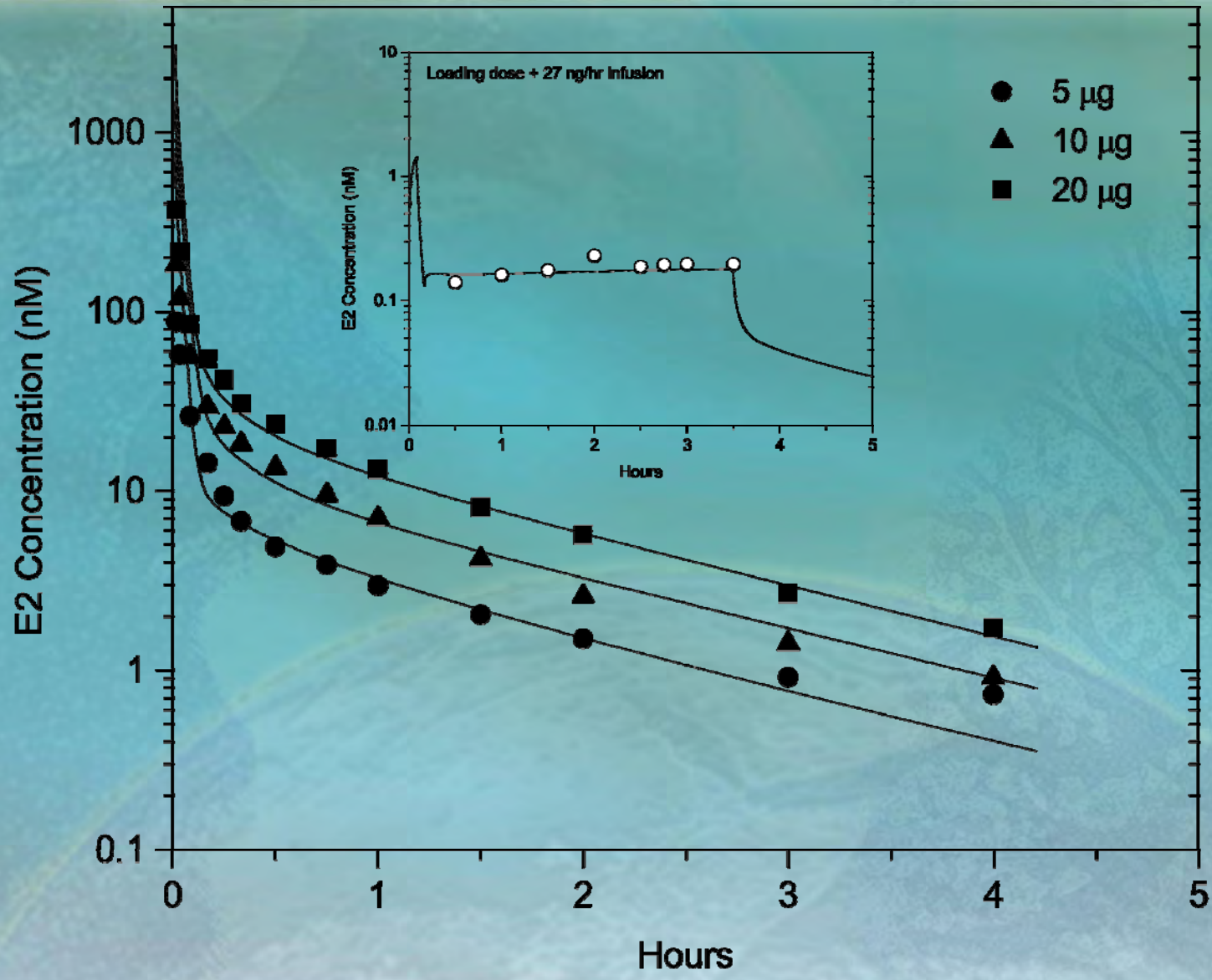
- IV E2 pharmacokinetics in intact female rats. ●, plasma; ■, uterus; ▼, liver. Dotted line: Diffusion limited uterine uptake

E2 Pharmacokinetics in OVX Female Rats



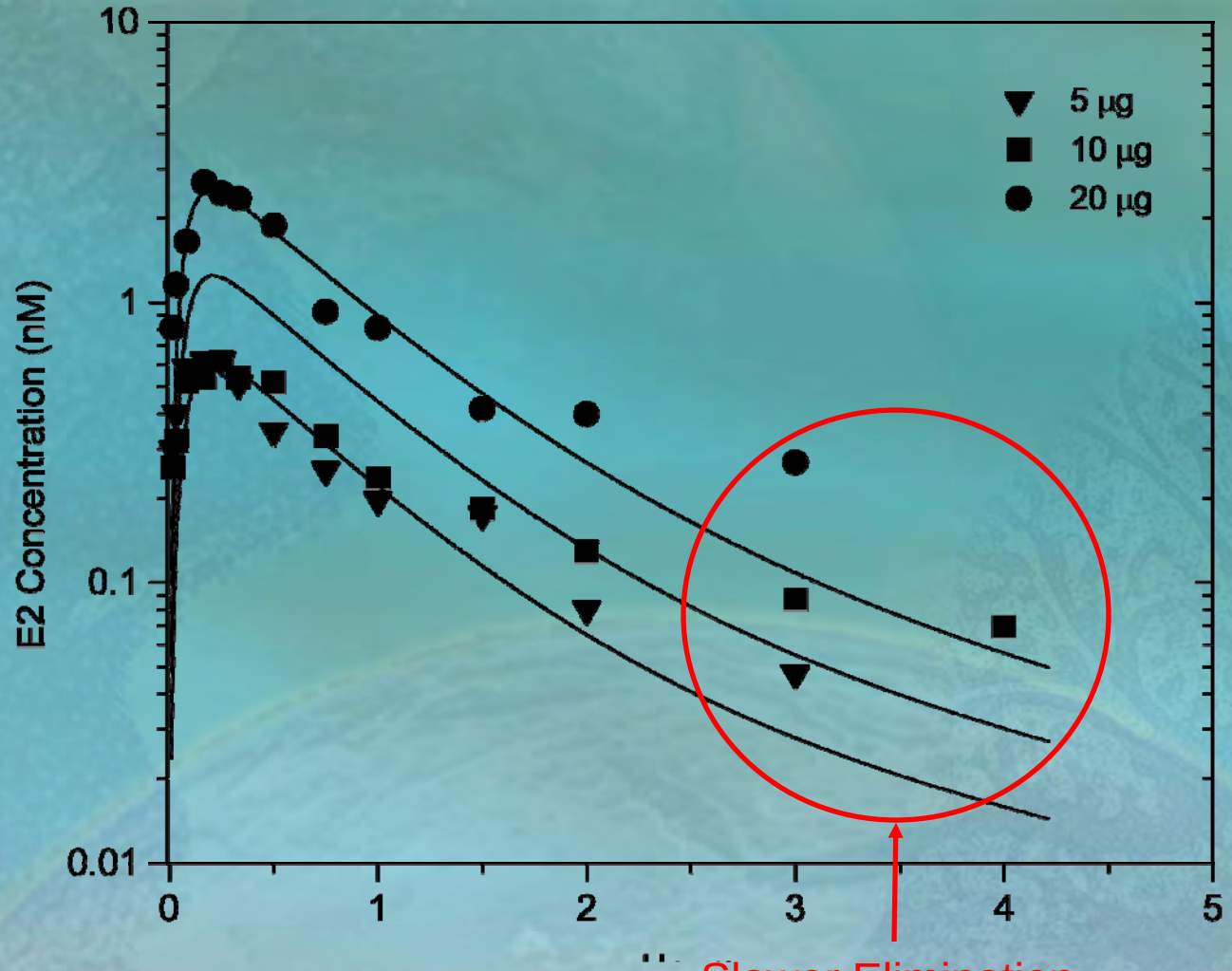
- IV E2 pharmacokinetics in OVX female rats. 2.5 (closed circles) and 25 (open circles) µg E2/kg. Dotted line: purfusion limited uterine uptake.

IV Route E2 Pharmacokintics in Male Rats



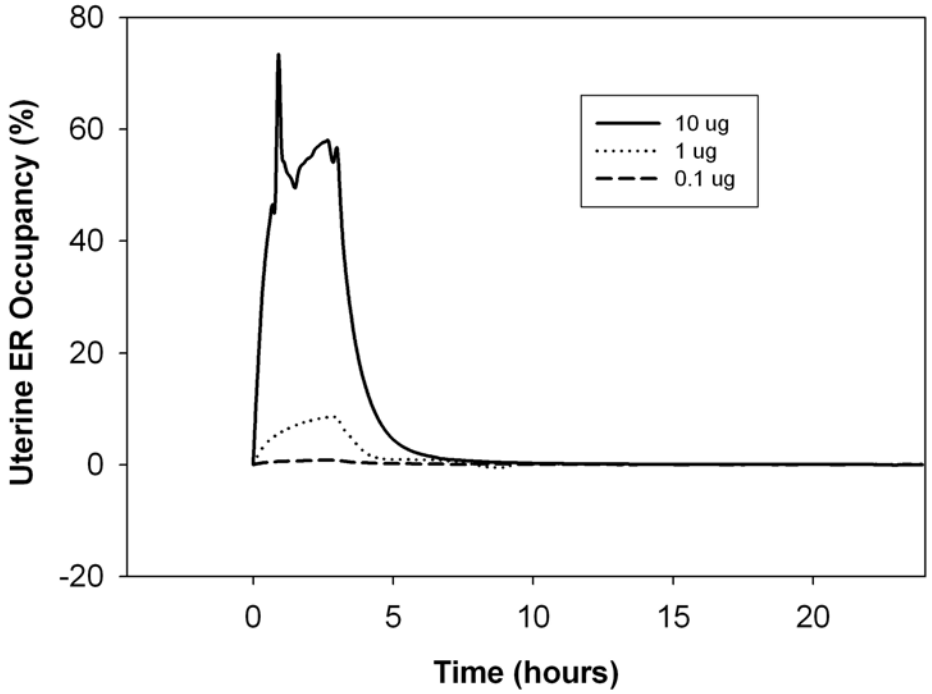
- IV E2 pharmacokinetics in male rats.

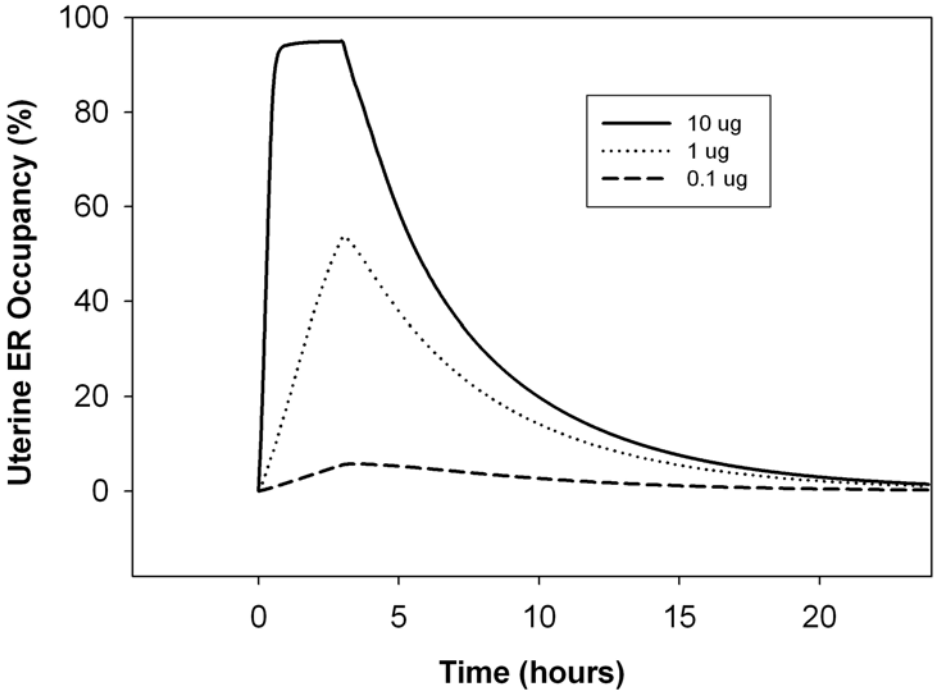
E2 Pharmacokintics
Intraduodenal Administration in Male Rats



Slower Elimination
Or
Eterohepatic Recirculation?

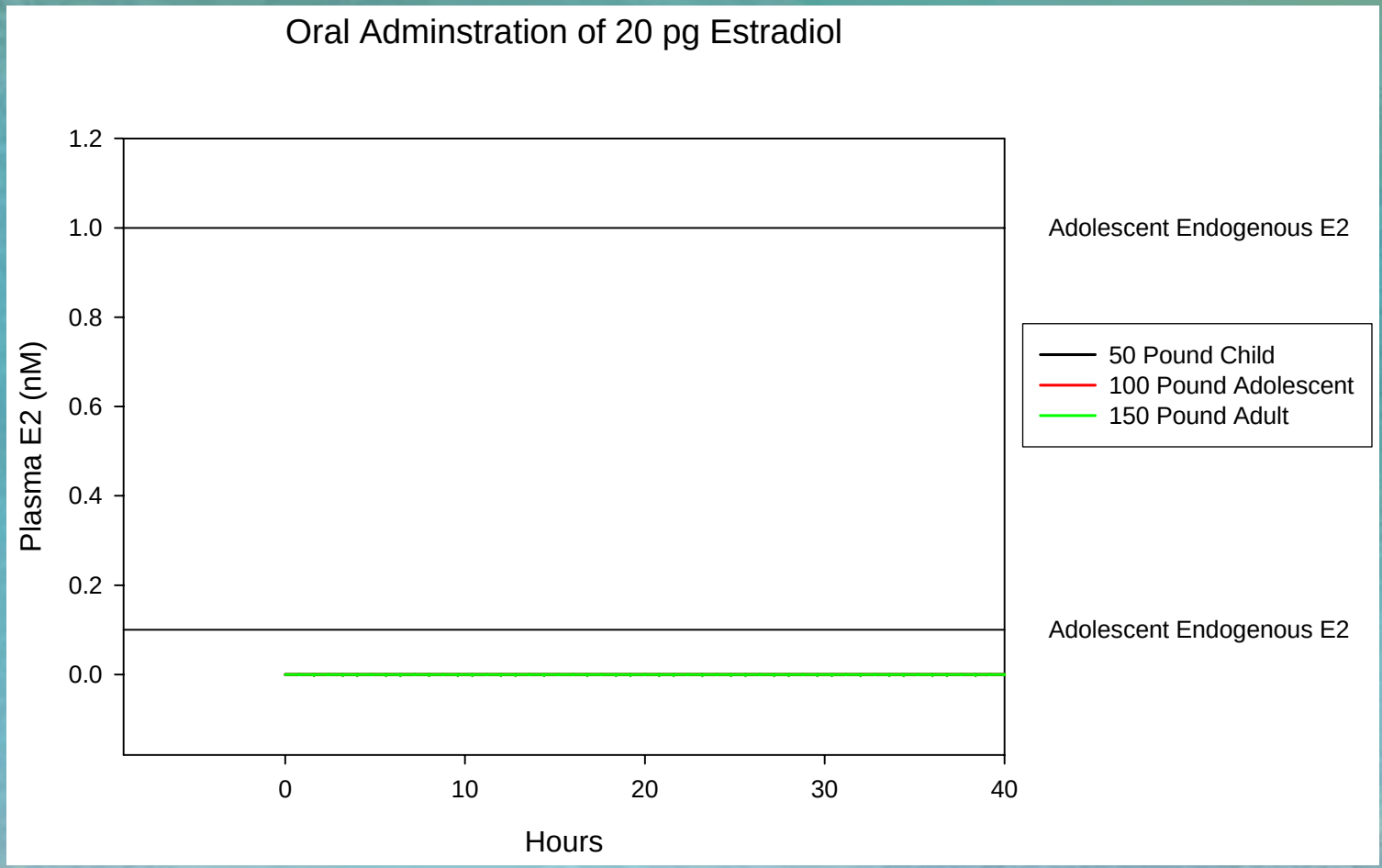
ER Binding and Uterine Response





- Subcutaneous E2, intact rats (Naciff et al, 2003)
- Uterine response
  - 0.1 µg/kg/day—100% of control
  - 1 µg/kg/day——380% of control
  - 10 µg/kg/day—430% of control

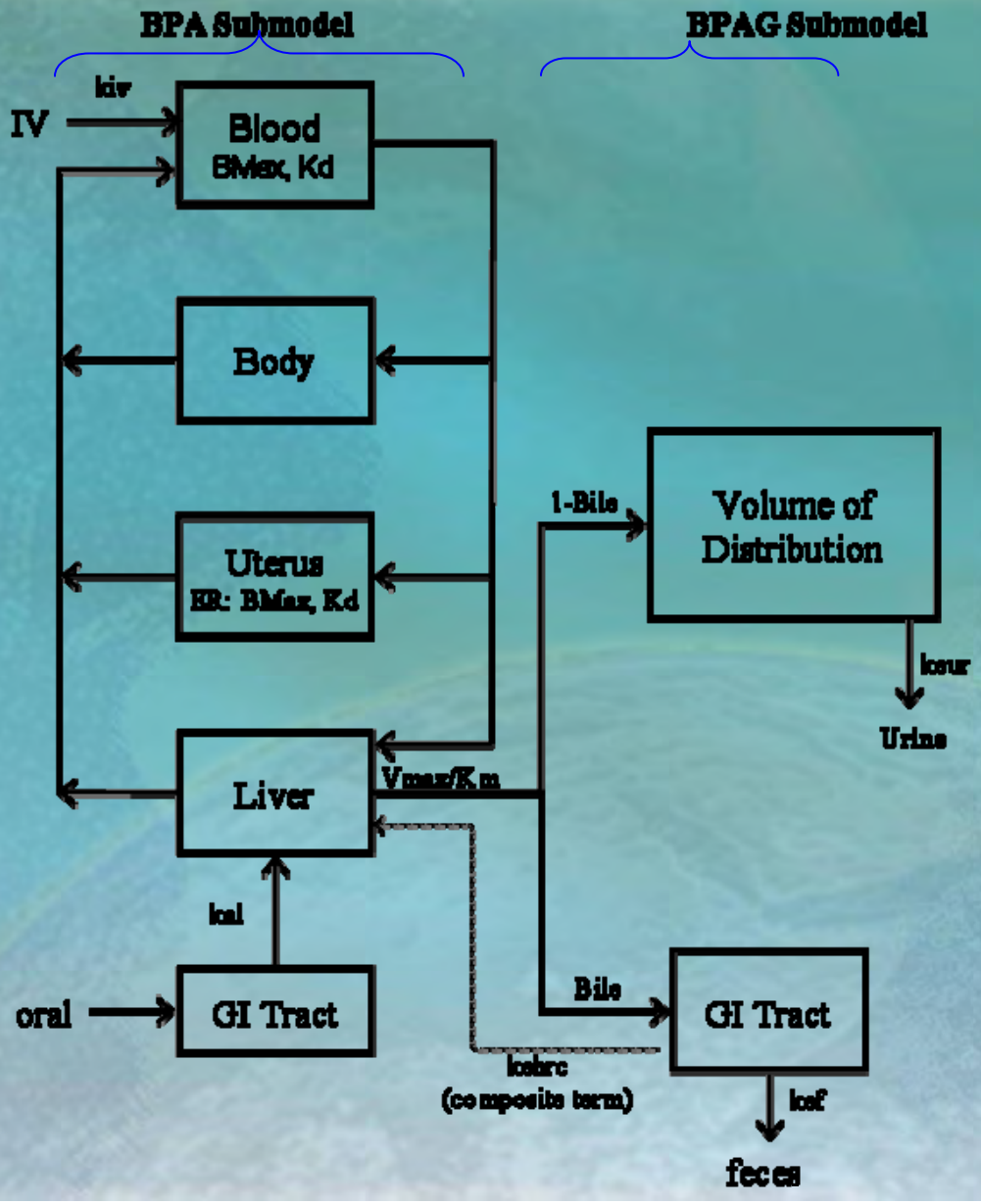
Does Estradiol and Estrone In Milk Pose a Risk to Adolescents?



10 pg of E2 and 10 pg of Estrone administered as E2

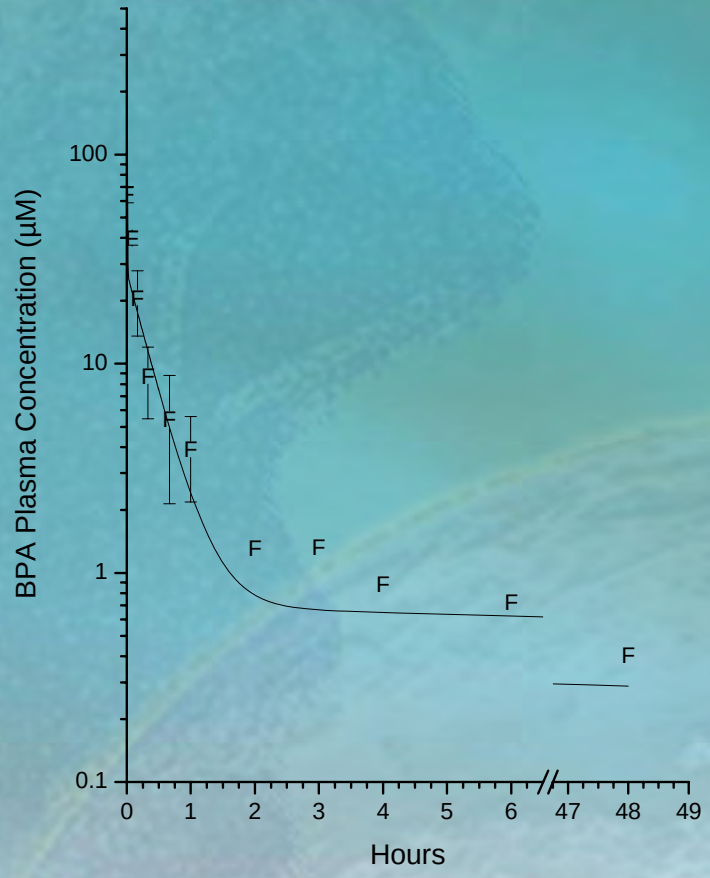
Bisphenol A

Bisphenol A Model

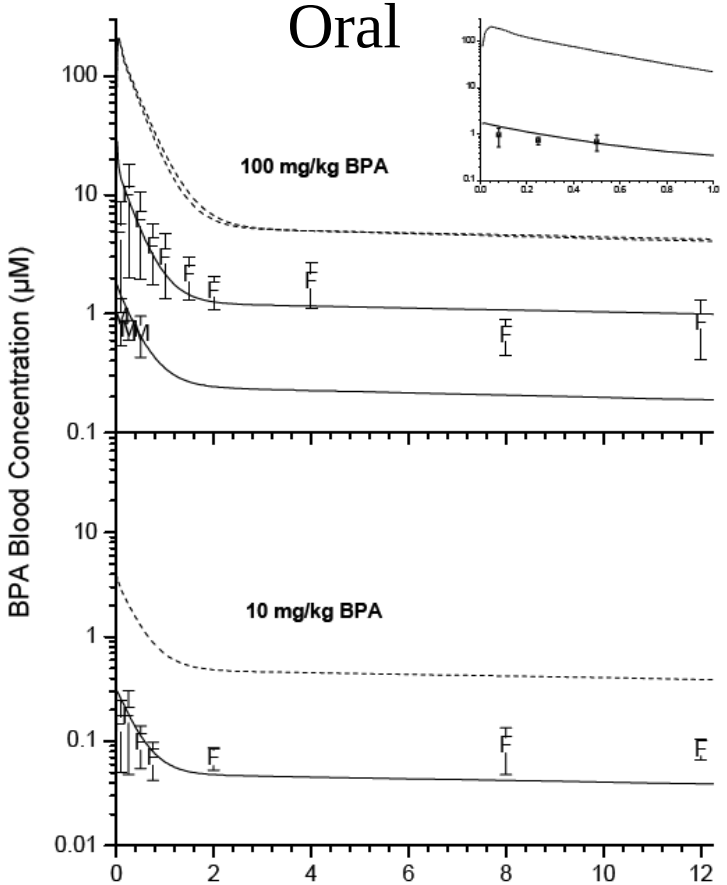


Rat Blood Kinetics

IV



Oral



Tissue Response is Related to Receptor Occupancy

Correlating Uterine Receptor Dose Metrics And Uterine Response

Oral Dose (mg/kg)	Percent Occupancy	Percent Occupancy	Uterine Weight (% control)[a]
	Without Binding	With Binding	
0	0	0	100
10	9.6	0.63	100
100	73.1	14.0	100
200	84.8	25.2	124
800	96.4	62	200
	ER-BPA Complex AUC Without Binding	ER-BPA Complex AUC With Binding	
0	0	0	100
10	0.025	0.002	100
100	0.19	0.04	100
200	0.23	0.07	124
800	0.26	0.17	200

a Twomey, 1998

- Response is not related to oral dose
- Response is not related to receptor occupancy in the absence of protein binding
- Response is best related to receptor occupancy when protein binding is considered

Conclusions

- **Protein binding can influence the biologically active fraction of a chemical in blood or a tissue**
  - **Critical for many pharmaceuticals**
  - **Affect cross species, sex and life-stage extrapolations**
- **Modeling at the PK/PD interface reveals important dose response relationships—at lease for BPA—so they are impactful**
- **Models like these are reasonably easy to develop and parameterize.**

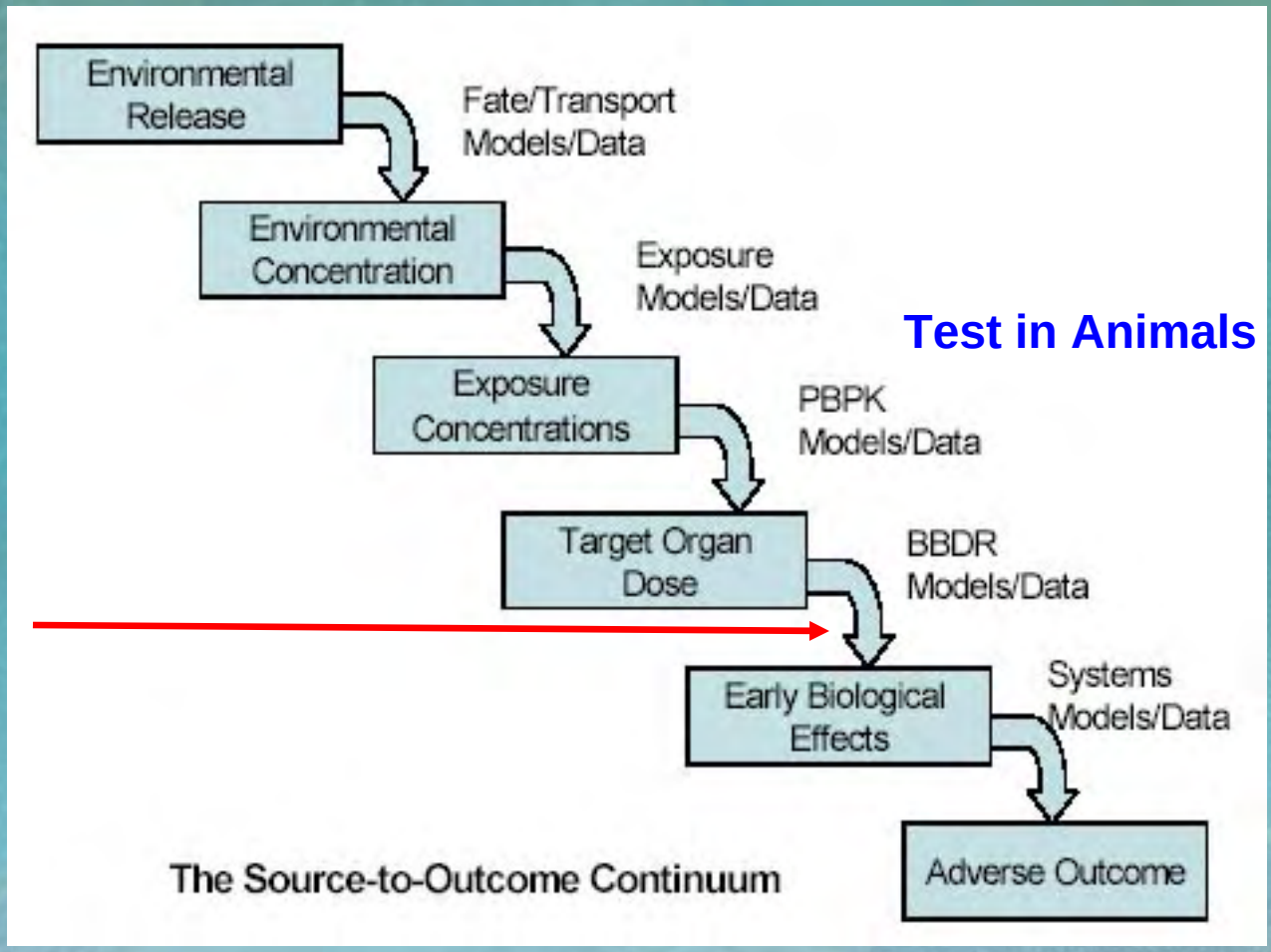
Acknowledgements

- Hugh Barton, EPA
- David Plowchalk, Pfizer
- American Chemistry Council and the American Plastics Council

Model This

Its Not About Modeling!

Why Model Receptor Binding?



Biologically Active Dose

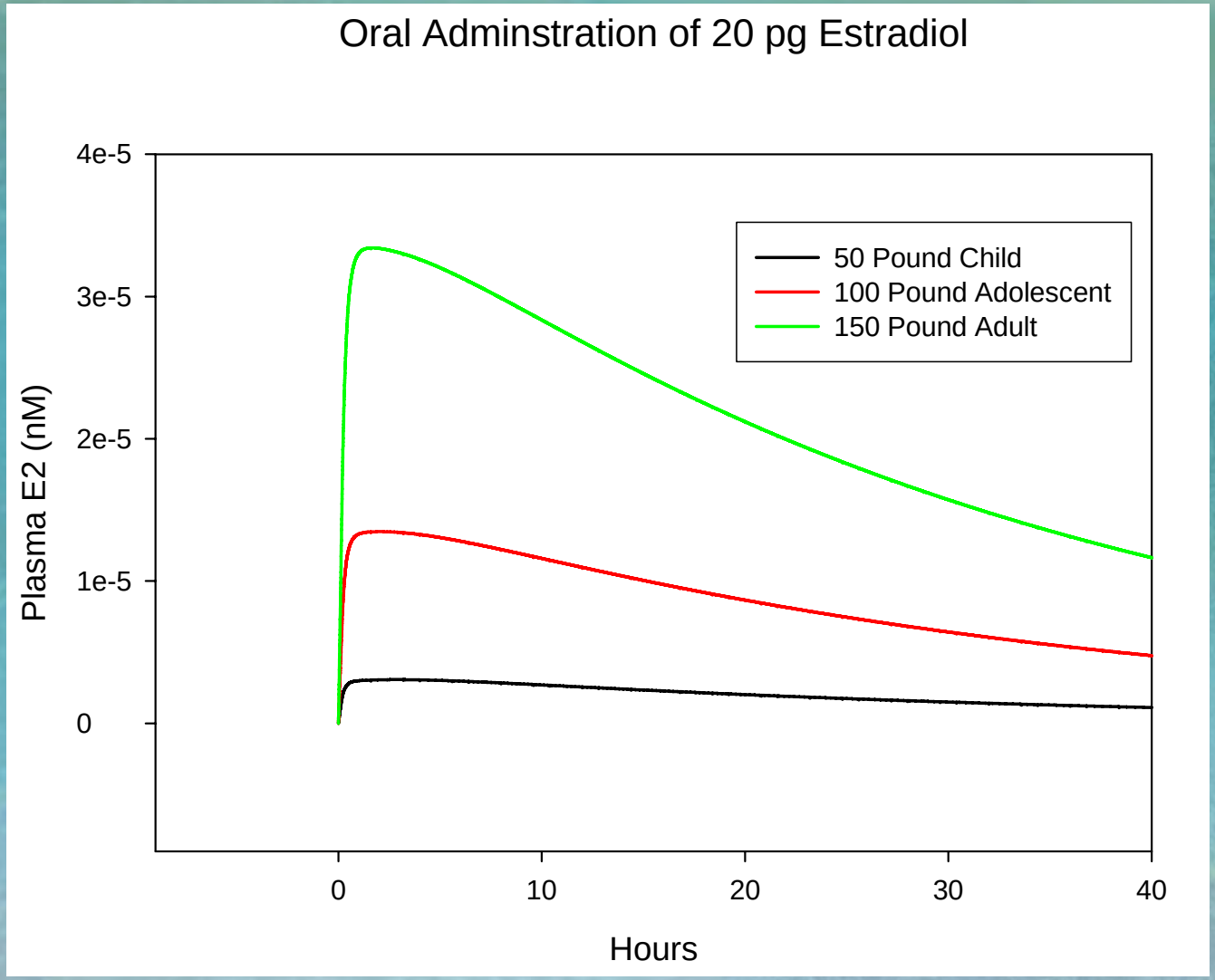
Test in Animals

Predict in Humans

What Exposure

Is SAFE?

Does Estradiol and Estrone In Milk Pose a Risk to Adolescents?



10 pg of E2 and 10 pg of Estrone administered as E2

Clearance Affects Potency

Potency Relative to E2

	In Vitro		*In Vivo*
Compound	Serum Free	Adult Rat Serum	Adult Rat
Estradiol	1	1	1
Genistein	0.02	0.16	0.32
Bisphenol A	0.0005	0.0009	0.002
Weak Agonist – Slow clearance	0.0005	0.0009	0.02
Weak Agonist – Fast clearance	0.0005	0.0009	0.0002

Adapted from: Teeguarden, J. G., and Barton, H. A. (2004). *Risk Anal* **24**, 751-70.

Simple Examples of the Importance of Binding and Clearance on Measures of Potency

Estradiol and Bisphenol A

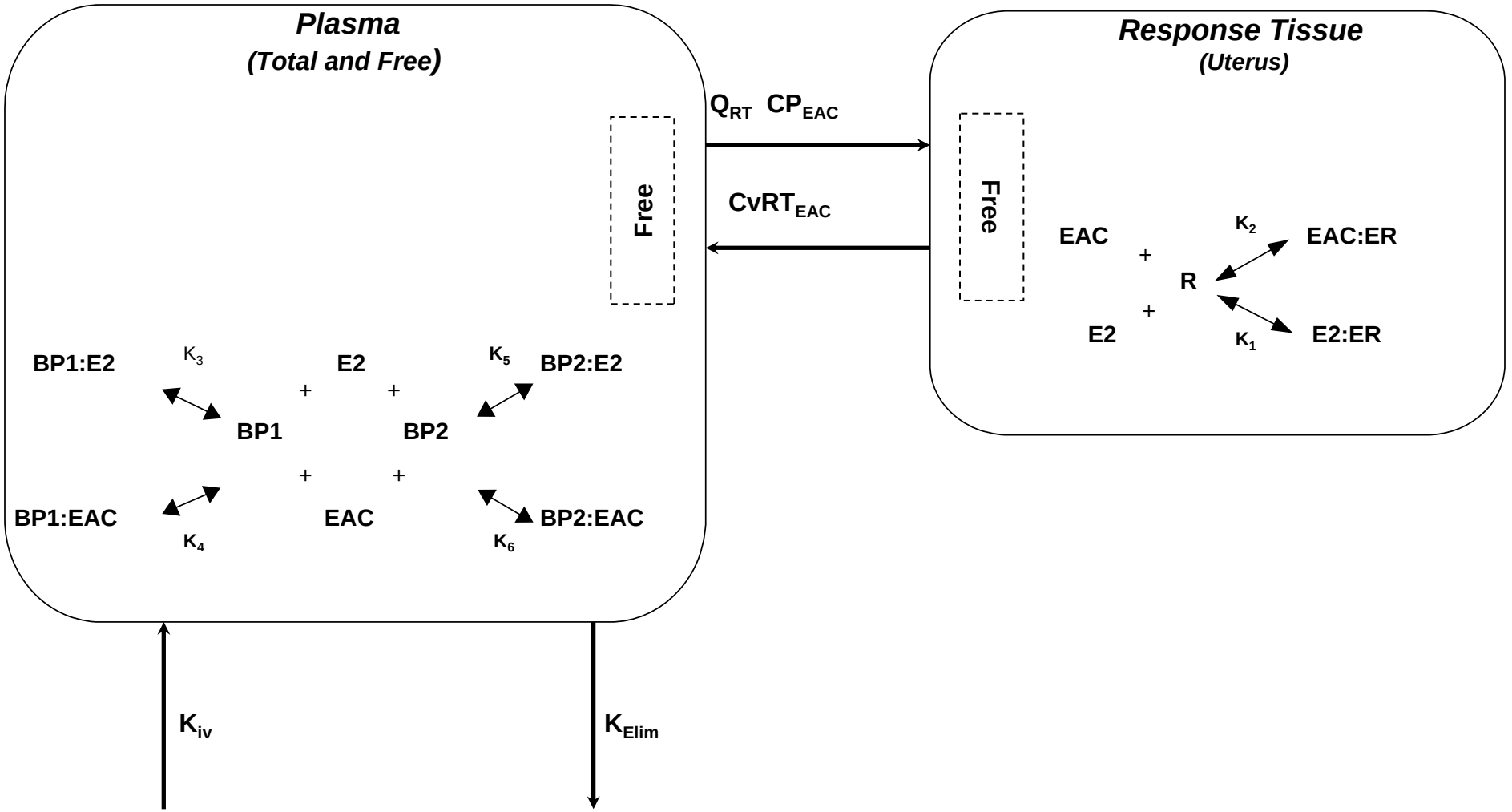
- **Estradiol**
  - **~ 2% non-bound (free) in human blood**
  - **Significant binding to SHBG and albumin**
  - **~ 4% free in rat blood, less during pregnancy**
  - **Significant binding to α-fetoprotein and albumin**
- **BPA**
  - **~ 7 % free in human blood (mostly albumin binding)**
  - **~ 6% free in rat blood (mostly albumin binding)**

Mendel, C.M. (1992) The Free Hormone Hypothesis: Distinction from the Free Hormone Transport Hypothesis. Journal of Andrology. 13(2): 107-116

Bisphenol A

- Weakly estrogenic plasticizing compound
- Acts through the estrogen receptor
- Highly bound in the blood
- Dose-response for rodents:
  - Affected by non linear kinetics?
  - Affected by protein binding in blood?
  - Is response related to receptor binding?
- Uterotrophic response: Linking receptor binding, tissue dynamics and a common reproductive/developmental toxicology endpoint

Integrating Protein Binding, Receptor Binding and Clearance Using *In Vivo* Models



Basic Principles: Hormone Transport

- The Free Hormone Transport Hypothesis
  - Only free hormone can cross cell membranes (from blood to tissues)
  - Tissue receptors can be in equilibrium with free and (sometimes) some portion of the protein bound
  - This means the extent and affinity of binding in blood is an important determinant of biological activity

Mendel, C.M. (1992) The Free Hormone Hypothesis: Distinction from the Free Hormone Transport Hypothesis. Journal of Andrology. 13(2): 107-116